Adaptation Strategies for Wild Blueberry Growers in a Changing Climate: Mulching Effects on Crop Productivity and Fertility Effects on Blueberry Gall Midge

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ADAPTATION STRATEGIES FOR WILD BLUEBERRY GROWERS IN A CHANGING CLIMATE: MULCHING EFFECTS ON CROP PRODUCTIVITY AND FERTILITY EFFECTS ON BLUEBERRY GALL MIDGE

By

Rebecca Gumbrewicz

B.S. University of Rhode Island, 2019

A THESIS
Submitted in Partial Fulfillment of the Requirements for the Degree of Master of Science (in Plant, Soil, and Environmental Sciences)

The Graduate School
The University of Maine
December 2021

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By Rebecca Gumbrewicz

Thesis Advisor: Dr. Lily Calderwood

An Abstract of the Thesis Presented in Partial Fulfillment of the Requirements for the Degree of Master of Science (in Plant, Soil, and Environmental Sciences) December 2021

Wild blueberry (Vaccinium angustifolium Aiton.) cropping systems are considered resilient to environmental changes due to ecological and genetic diversity within each field. However, wild blueberries can be sensitive to weather fluctuations that cause extreme temperature or moisture regimes. Climate change in Maine is represented by increasing rates of warming temperatures, more intense precipitation events, and more frequent atmospheric “blocking” patterns. Warming temperatures result in the northward expansion of pest ranges and altered growing seasons. More extreme rainfall events lead to damaged plantings and soil erosion. Atmospheric blocking leads to an increased likelihood of heat waves and drought. Two experiments were conducted over one production cycle to identify potential management strategies that can help wild blueberry growers mitigate drought and pest challenges in Maine.

The purpose of the first experiment was to determine the effect of wood mulch particle size on wild blueberry soil, plant growth, pest pressure and yield. Four mulch treatments representing four different particle sizes were applied at a thickness of 1.27 cm in an organic wild blueberry field in Stockton Springs, ME. Treatments from smallest to largest were sawdust, shavings, bark mulch and wood chips. No mulch treatment retained significantly more soil
moisture than the control. All mulch treatments significantly reduced disease compared to the control in year one. Sawdust and shaving treatments, the two smallest particle sizes, resulted in the greatest yield. These results indicated that wild blueberry growers should consider smaller particle sizes rather than the traditional wood chip mulch when choosing a mulch type to apply.

The purpose of the second experiment was to measure the effect of diammonium phosphate (DAP) fertilizer application on a more recent pest of wild blueberry, the blueberry gall midge (*Dasineura oxycoccana* Johnson). This pest has been observed in high densities where there were high soil and foliar nutrient levels in both wild blueberry and cranberry (*Vaccinium macrocarpon* Aiton.) systems. This study was conducted in two conventional wild blueberry fields in Jonesboro and Washington, ME. The effects of treatment combinations of DAP application with galling on wild blueberry soils, plant growth and yield were recorded. Gall density was significantly greater in fertilized plots during the prune year in Jonesboro and both years in Washington. Foliar percent nitrogen and phosphorus had a significant positive linear relationship with gall density. Stems with galls and without DAP applied were significantly shorter and had fewer buds per stem compared to fertilized stems without galls at both sites. Stems without galls and with DAP applied had the greatest number of buds, flowers, and berries per stem at the Washington site. Mean stem height and total yield at this site were greater in fertilizer treatments even when galling was present. It is important for growers applying fertilizers to monitor field infestation levels due to our findings that DAP fertilizer impacted the economic thresholds for this pest and DAP fertilizer combined with galling impacted blueberry development and yield. Both studies provide essential information for wild blueberry growers to make more informed management decisions when the effects of climate change make such decisions increasingly difficult as weather trends become more unpredictable.
DEDICATION

This thesis is dedicated to my beloved family, friends, God Almighty, and Jesus Christ my Lord and Savior. Their constant support and encouragement have given me the strength and knowledge to complete this work. Thank you to my advisor and committee members, whose guidance and critique helped make this work possible. Thank you to my undergraduate professors who gave me the experience and motivation to pursue an endeavor like this. Thank you to my parents, whose love and support through things big and small means the world. God bless.
ACKNOWLEDGEMENTS

I thank University of Maine undergraduates Sydney Abramovich, Abby Cadorette, Erica Carpenter, and Aidan Lurgio for their valued hard work and assistance with data collection and entry. We also thank graduate students Anthony Ayers and Pratima Pahadi, and lab technicians Brogan Tooley and Mara Scallon for their teamwork and advice both with data collection and during the writing stages of this thesis. We are grateful to University of Maine faculty members Judith Collins, Frank Drummond, Phil Fanning, and Bill Halteman for their support and guidance during the planning, analysis, and writing stages of this study. We also want to emphasize our thanks to the wild blueberry growers for their continuous support, feedback, and permission to use their land to conduct our research. This work was supported by the Northeast Sustainable Agriculture Research and Education (SARE) program under grant number LNE19-374, the Maine Food and Agriculture Center, and the Wild Blueberry Commission of Maine.
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CHAPTER 1: EXPLORING WOOD MULCH PARTICLE SIZE AS A DROUGHT BUFFER AND ORGANIC MATTER BUILDER IN WILD BLUEBERRY (VACCINIUM ANGUSTIFOLIUM AITON.)

1.1 Chapter Abstract
Climate change in Maine is characterized by increasing rates of warming temperatures and more intense rain events, which lead to altered growing seasons, earlier emergence of pests, increased seasonal drought and, in turn, large crop losses. These changes have significant impacts on wild blueberry systems and make it more difficult for growers to manage this crop under increasingly unpredictable weather patterns. Mulching is a tool wild blueberry growers use to combat some of these challenges, but the comparative effects of wood mulch particle size in wild blueberry systems has not, to our knowledge, been studied. The purpose of this study was to determine the effect of wood mulch particle size on wild blueberry soil, plant development, pest pressure and yield. This study was carried out over a two-year period (one production cycle) in an organic wild blueberry field in Stockton Springs, ME. No mulch treatment demonstrated significantly greater overall mean soil moisture compared to the control at the 1.27 cm application thickness. All mulch treatments significantly reduced disease pressure, primarily Sphaerulina leaf spot caused by *Sphaerulina vaccinii*, compared to the control in year one. The two finest particle size treatments, sawdust and shavings, resulted in significantly greater yields than the control, and were the least expensive material. This suggests that growers should consider using finer particle size mulches rather than the current tradition, wood chips, but may need to reapply at a more frequent rate. Any mulch particle size application is better than no application.
1.2 Introduction

1.2.1 Wild Blueberry Production

Wild blueberries (Vaccinium angustifolium Aiton.) are a native species and culturally and economically significant perennial fruit crop in the northeastern United States and Canada. Maine and eastern Canada are the world’s leading wild blueberry growing regions where nearly 22,000 and 82,000 metric tons, respectively, were produced in 2021 (IBO, 2021). Growers manage commercial wild blueberry fields on a two-year production cycle. In year one (prune year), fields are mowed or burned to stimulate new vegetative growth, resulting in flower buds. In year two (crop year), plants bloom and berries develop for harvest in August. The two-year cropping cycle is used as a pest and crop management tool, specifically to reduce weed, insect pest, and plant pathogen populations in addition to promoting new shoot and bud development for increased yield and quality (Drummond et al. 2009).

1.2.2 Climate Change Impacts on Wild Blueberry

The Northeast is warming faster than any other region in the U.S. (Fernandez et al. 2020). The average annual temperature in Maine as of 2020 has increased by 1.78°C since 1895 with the greatest temperature increases occurring along the coast, where most wild blueberries are grown in Maine (Fernandez et al. 2020; Tasnim et al. 2021). Previous research suggests hotter temperatures may cause earlier emergence of insect pests, such as spotted wing Drosophila, Drosophila suzukii (Matsumura) (Diptera: Drosophilidae) (Drummond et al. 2019). Warming temperatures also lead to worsening periods of drought as they favor drying and amplify soil moisture deficits (Gu et al. 2019; Naumann et al. 2018; Samaniego et al. 2018). Sufficient soil moisture is necessary for optimal vegetative growth, bud development, fruit ripening, and yield for many small fruit crops, including wild blueberries (Davies and Albrigo, 1983; DeGomez and
Wild blueberries are grown in sandy, well-draining soils that have a low water holding capacity and are therefore unable to maintain adequate soil moisture for long periods of time. Without sufficient soil moisture, plants cannot absorb the nutrients they need for leaf, bud, and berry development. Increasing soil moisture allows more nutrients to dissolve in solution, resulting in a greater total number and weight of berries (Benoit et al. 1984) while drought conditions result in undersized fruit, reduced stem water potential, and lower transpiration and photosynthetic rates (Glass et al. 2003; Hicklenton et al. 2000). Heat waves and drought become even more likely with the stagnation of weather patterns and more frequent atmospheric “blocking” (Birkel and Mayewski, 2018). According to the U.S. Drought Monitor, all but four years since 2000 in Maine have exhibited abnormally dry conditions during the summer months of June, July, and August (U.S. Drought Portal, 2020), critical growing months in both prune and crop years for wild blueberry. Drought conditions in 2020 resulted in 43.7% mean crop loss (Schattman et al. 2021).

Average annual precipitation has increased by 14.73 cm (15%) since 1895 (Fernandez et al. 2021), along with the intensity of such rain events, which lead to damaged plantings and increased soil erosion (Birkel and Mayewski, 2018). Increased spring rainfall has led to a decline in pollination days, days that pollinators will forage during bloom, which has potential for further crop losses (Drummond et al. 2017). This increased precipitation is also associated with September and October tropical cyclones, as well as warming temperatures in the Atlantic Ocean (Frei et al. 2015; Huang et al. 2018; Huang et al. 2017). Therefore, the timing of these events does not always occur at ideal points during the wild blueberry growing season when plants would take up the most water.
This crop requires 2.54 cm of water each week from April through October to meet plant demand (Trevett, 1967; Hunt et al. 2008). Rainfall history from 1959 to 1998 across the Maine wild blueberry growing region shows the probability of reaching this rainfall requirement each week is less than 50% throughout the growing season, and less than 20% during the critical fruiting period, mid-July through August (Dalton and Yarborough, 2004). Long term weather trends estimate that wild blueberries will lack sufficient water in August in four of every five years (Hunt et al. 2009).

1.2.3 Mulching Wild Blueberry

Mulch can be any material spread on top of the soil, rather than incorporated into it (Chalker-Scott, 2007). Mulch can be used for several purposes, including reduction of water loss, moderation of soil temperature, suppression of weed growth, prevention of disease spread, and stimulation of rhizome growth (Drummond et al. 2009; Yarborough, 2012). Chalker-Scott (2007) notes that organic mulches are best-suited for achieving overall plant performance compared to inorganic or synthetic materials like gravel or plastic mulches. Mulching with organic matter covers the soil surface, preventing it from drying out, sealing shut, and losing porosity (Bot and Benites, 2005). Ultimately, mulching with organic matter helps promote a more drought-resistant soil.

Mulching and mowing are the primary methods that wild blueberry farmers use to build organic matter, thereby improving soil health. Biannual pruning of wild blueberry fields by mowing increases the thickness of the “organic pad” as plant debris decomposes on the soil surface. This O horizon, or “organic pad”, serves as a substrate with high water holding capacity due to high levels of organic matter between 11-12% (Argall et al. 1998; Eaton et al. 2009). Organic growers use prescribed burning to double as a pruning and pest management tactic yet
burning for long periods of time or at very hot temperatures can reduce the amount of valuable organic matter on the soil surface. An insufficient organic layer at the soil surface will allow leaching of necessary soil nutrients, resulting in a loss of any added nutrients from the blueberry root zone. Increasing soil organic matter can increase water holding capacity and result in greater retention of soil moisture, allowing more nutrients to dissolve into the soil solution. This makes essential nutrients more readily available for plant uptake, especially when the acidic pH of wild blueberry soils (4.0-5.0) makes the availability of macronutrients like nitrogen and phosphorous even more limited (Jones and Jacobsen, 2005). The abundance of soft wood materials in Maine coupled with ease of spreading material by manure spreaders across fields makes mulching a viable cultural management option for farmers interested in increasing soil water availability by reducing water loss and increasing organic matter rather than increasing water inputs.

On Maine wild blueberry farms, wood mulch, usually in the form of wood chips, has traditionally been applied in thick patches between five and ten centimeters deep to cover bare spots, encourage rhizome expansion, and prevent weed growth (DeGomez and Smagula, 1990b; NRCS 2017). Mulching is an effective tool to prevent spore dispersal and has been shown to significantly reduce mummy berry blight in wild blueberry (Drummond et al. 2009). Smagula and Goltz (1988) showed that mulching areas of wild blueberry fields enabled rapid growth of blueberry seedlings, while unmulched areas experienced minimal survival due to frost heaving. Hunt et al. (2010) found that pine bark mulch significantly improves soil water retention, with the greatest effects observed during dry periods but still effective during periods of excessive rainfall. Sanderson and Cutcliffe (1991) found that a sawdust mulch applied at a thickness of 5 cm increased total wild blueberry yield by nearly 30% when compared to 0 cm and 10 cm
treatments. However, research on the comparative effects of mulch particle size has not, to our knowledge, been investigated in the wild blueberry cropping system.

Due to the lack of research on mulch particle size effects in wild blueberry this study seeks to evaluate the effects of varying particle sizes of organic pine mulch on wild blueberry growth and soil characteristics. Specifically, our objectives were to determine the effects of mulch particle size on (i) the wild blueberry soil environment (soil moisture, temperature, nutrients, and organic matter), (ii) wild blueberry plant growth (vegetative and reproductive growth i.e., foliar nutrients, stem height, bud/flower/fruit development) and (iii) pest pressure (density and cover of weeds, insects, and disease). We hypothesized that mulch treatments would increase soil moisture, soil organic matter, plant growth and yield as well as reduce pest pressure compared to the control treatment. Specifically, we hypothesized that finer particle sizes would be more effective in increasing soil moisture by contributing a greater fraction to soil organic matter as they decomposed compared to larger particle sizes.

1.3 Materials and Methods

1.3.1 Study Site

The study took place in a MOFGA (Maine Organic Farmers and Gardeners Association) certified organic field in Stockton Springs, ME, USA. The field was in the prune year in 2020 and crop year in 2021. The experiment began with mulch application in May 2020 and continued through September 2021 (one production cycle). The experimental design is a randomized complete block design with six replicates. Each plot was 1.83 m by 9.14 m (16.72 m²) separated by 0.91 m buffers.
1.3.2 Treatments

Wild blueberry plant growth and soil characteristics were measured under five treatments including four different mulch particle sizes and a control (no mulch). Particle size was determined using 2.0-, 4.0-, 6.3-, 9.5-, and 12.5-mm soil sieves. The fraction of material able to pass through each sieve was weighed to calculate the percent of material within each sieve size range (Table 1.1). The median particle size ($D_{50}$), the sieve size that allows 50% of particles to pass through, was used to rank mulch treatments by the most abundant particle size in each (Corey and Kemper, 1968; Diaz et al. 2005). Mulch treatments are ranked by increasing size as follows: sawdust (smallest, $D_{50} < 2\text{mm}$), wood shavings (medium, $2.0 \text{ mm} < D_{50} < 4.0 \text{ mm}$), bark mulch (large, $4.0 \text{ mm} < D_{50} < 6.3 \text{ mm}$), and wood chips (extra-large, $6.3 \text{ mm} < D_{50} < 9.5 \text{ mm}$).

A sample of each mulch particle size material was collected on May 20, 2020 and submitted to the Maine Agricultural and Forest Experiment Station Analytical Laboratory for analysis of pH, nitrogen, calcium, potassium, magnesium, phosphorus, aluminium, boron, copper, iron, manganese, and zinc. A separate sample of each mulch type was weighed before and after drying in a gravity convection oven (Thermo Scientific PR305225G, Columbia, MD, USA) for 48 hours to determine the percent moisture of each material (Table 1.2). Mulches were all from the same material, untreated white pine ($Pinus strobus$), purchased as by-products from N.C. Hunt Lumber, a sawmill and manufacturing company in Jefferson, Maine. All products were applied once during the prune cycle on May 19 and 20, 2020 at a thickness of 1.27 cm.
Table 1.1. Particle size distribution for each mulch type by percent of dry matter within each sieve size range.

<table>
<thead>
<tr>
<th>Mulch type</th>
<th>Particle size distribution (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&lt;2.0</td>
</tr>
<tr>
<td>Sawdust</td>
<td>72.68</td>
</tr>
<tr>
<td>Shavings</td>
<td>36.36</td>
</tr>
<tr>
<td>Bark</td>
<td>12.79</td>
</tr>
<tr>
<td>Chips</td>
<td>0.30</td>
</tr>
</tbody>
</table>
Table 1.2. Mulch treatment characteristics (pH, chemical composition, mean moisture content, dry matter application rate and cost). Prices may vary based on retailer and quantity purchased. Estimates do not include labor costs. Prices are based on one application of each material applied at a 1.27 cm thickness.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>pH</th>
<th>N (%)</th>
<th>Ca (%)</th>
<th>K (%)</th>
<th>Mg (%)</th>
<th>P (%)</th>
<th>Al (mg/kg)</th>
<th>B (mg/kg)</th>
<th>Cu (mg/kg)</th>
<th>Fe (mg/kg)</th>
<th>Mn (mg/kg)</th>
<th>Zn (mg/kg)</th>
<th>Moisture (%)</th>
<th>Dry Matter Rate (kg/ha)</th>
<th>Cost ($/m³)</th>
<th>Cost ($/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sawdust</td>
<td>4.9</td>
<td>0.050</td>
<td>0.076</td>
<td>0.049</td>
<td>0.014</td>
<td>0.006</td>
<td>67.2</td>
<td>2.68</td>
<td>5.32</td>
<td>111.0</td>
<td>70.3</td>
<td>7.06</td>
<td>52.52</td>
<td>12,976</td>
<td>12.97</td>
<td>1647.19</td>
</tr>
<tr>
<td>Shavings</td>
<td>5.1</td>
<td>0.049</td>
<td>0.040</td>
<td>0.027</td>
<td>0.012</td>
<td>0.002</td>
<td>32.5</td>
<td>2.01</td>
<td>3.83</td>
<td>67.6</td>
<td>29.2</td>
<td>7.76</td>
<td>28.41</td>
<td>4,363</td>
<td>12.97</td>
<td>1647.19</td>
</tr>
<tr>
<td>Bark Mulch</td>
<td>5.3</td>
<td>0.154</td>
<td>0.199</td>
<td>0.078</td>
<td>0.025</td>
<td>0.016</td>
<td>173.0</td>
<td>5.59</td>
<td>2.94</td>
<td>157.0</td>
<td>69.0</td>
<td>23.1</td>
<td>58.65</td>
<td>11,005</td>
<td>37.28</td>
<td>4734.56</td>
</tr>
<tr>
<td>Wood Chips</td>
<td>6.1</td>
<td>0.060</td>
<td>0.068</td>
<td>0.086</td>
<td>0.014</td>
<td>0.011</td>
<td>16.4</td>
<td>1.97</td>
<td>2.27</td>
<td>19.9</td>
<td>72.3</td>
<td>&lt;2.46</td>
<td>56.95</td>
<td>14,788</td>
<td>39.89</td>
<td>5066.03</td>
</tr>
</tbody>
</table>
1.3.3 Data Collection

1.3.3.1 Soil Environment

Soil samples were taken using a hand-held soil probe to a depth of 10 cm before treatments were applied on May 19, 2020. Each plot was soil sampled again after harvest on July 30, 2021, the final year of the project. Any visible mulch material that mixed with the soil during sampling was removed from soil samples collected after harvest prior to analysis. All samples were analyzed using standard field soil tests by the Maine Agricultural and Forest Experiment Station Analytical Laboratory (Hoskins, 1997).

Soil temperature (°C) and moisture (% volumetric water content [VWC]) were monitored weekly using TDR-150 (time domain reflectometry) probes (FieldScout TDR 150 Soil Moisture Meter, Spectrum Technologies, Inc., Aurora, IL., USA) and inserting them 12 cm (4.8 in.) into the soil of the blueberry root zone. Eight random readings were recorded per plot between 10:00 am and 2:00 pm on each collection date to maintain relatively consistent mid-day sampling conditions. Collection dates were performed weekly and, when possible, twice weekly before and after rain events. Collection dates for 2020 were June 2, 4, 8, 16; July 1, 10, 15, 24, 31; and August 10, 20, 31. Collection dates for 2021 were June 1, 10, 18, 24; and July 1, 6, 16, 22, 28.

1.3.3.2 Plant Growth

1.3.3.2.1 Prune Year Measures

Stem density was measured on June 19 and August 31, 2020 by counting the number of stems in each of two permanent 0.37-m² quadrats randomly placed in each plot at the beginning of the season and flagged for repeated use.

1.3.3.2.2 Crop Year Measures

Stem height and bud number per stem were recorded for each of five stems within each quadrat
on April 23, 2021. These five stems were marked with metal tags and used to measure fruit
development. The number of open flowers per stem was recorded during peak bloom on May 21,
2021. The number of green, red, and blue fruit per stem was recorded three times, once prior to
full ripening on June 18, 2021, once during ripening on July 6, 2021, and once during harvest on

Berries were harvested by hand raking plots on July 28, 2021. Quadrat yield was raked
and weighed separately before being added to the whole plot yield. Measures of berry quality
included 100-berry weights as an indication of berry size and °Brix as an indication of sugar
content. For 100-berry weights, a representative sample of 100 berries were randomly counted
and weighed. This sample was then blended using a Ninja Express Chop blender (SharkNinja
Operating LLC., Newton, MA, U.S.A). A subsample was deposited on a handheld PAL-
BRIX/ACID F5 refractometer (Atago, Saitama, Japan) for the brix measurements taken on July
30, 2021.

1.3.3.2.3 Both Year Measures

Blueberry cover was visually estimated per quadrat using a 0-6 scale ranking system with even
intervals that sum to 100%, where: 0 = not present, 1 = ≤1%-16.67%, 2 = >16.67%-33.33%, 3 =
>33.33%-50%, 4 = >50%-66.67%, 5 = >66.67%-83.33% and 6 = >83.33%-100%. Blueberry
cover was recorded in 2020 on June 19, July 15, and August 20 and in 2021 on June 18 and July
13.

Pest pressure was measured monthly using the same quadrats and ranking system used
for estimating blueberry cover. Each quadrat was evaluated for cover and density of weeds,
insects, and disease. Weed plants were recorded as broadleaf or grasses. Broadleaf plants were
identified to species while all grass species were grouped together. Diseases and insects were
identified to species. Pest pressure measurements were recorded on the same dates as for blueberry cover.

Twenty stems from each plot were randomly selected and marked with flagging tape to record leaf chlorophyll content in SPAD units monthly. This was measured as an indicator of leaf nitrogen (Xiong et al. 2015) using a SPAD Chlorophyll Meter (SPAD 502; Minolta Corp, Osaka, Japan). One measurement was taken on each of two leaves per stem, one on the lowermost portion and one on the uppermost portion. These measurements were recorded in 2020 on June 19, July 15, and August 20 and in 2021 on June 18 and July 16.

1.3.3.3 End of Data Collection

Wild blueberry plant development during 2021 was at least two weeks ahead of what was observed in 2020, which was also several weeks ahead in development compared to past years. Therefore, the harvest took place much earlier in the season. Any measurements planned for August 2021 were terminated after harvest.

1.3.4 Data Analysis

The effects of mulch treatments on soil (moisture, temperature, nutrients), pest pressure (insects, disease, weeds), plant growth (stem height, bud/flower/berry development, stem density, blueberry cover, chlorophyll content), yield and berry quality were statistically analyzed using JMP software (JMP® Pro, Version 15.2.0) (SAS Institute, 2020b). One-way analysis of variance was used to analyze data sets for only one date, including stem density, stem height, bud number per stem, flowers per stem, berries per stem, and soil nutrients. Randomization testing was used to analyze data that did not follow a normal distribution and confirm results of analyses of variance that did not meet standard parametric assumptions.
All repeated measures were analyzed using a mixed model. Data that met assumptions of normality and equal variances were analyzed using a linear mixed model of standard least squares. Data that could not meet assumptions of normality and equal variances were analyzed using a generalized linear mixed model (GLMM) after concluding there were no other major problems with the data set (SAS Institute, 2020a). In the GLMM, count data were modeled with a Poisson distribution and log link. Proportion data (ranks) were modeled with a binomial distribution and logit link. To model rank data, ranks were first converted to the percent midpoint of the range that each rank represents. In each model, ‘block’ was used as a random effect and all other variables were considered fixed effects. All fixed-by-fixed effect interactions were included in each model for statistical analysis. Treatment effects were separated by Tukey’s Highly Significant Differences test at the 0.05 significance level.

1.4 Results
The Maine statewide 2020 and 2021 summer (June – August) temperature and rainfall averages were warmer and drier than normal (Birkel, 2020a; 2021). The average summer temperature in 2020 was 18.72°C, the third warmest since 1895. The average summer temperature in 2021 was 18.67°C, the fourth warmest since 1895, following 2020. Total precipitation for the 2020 summer period was 22.35 cm, the 14th driest since 1895. Total precipitation for 2021 was 26.00 cm, 2.03 cm less than the historical mean.

1.4.1 Soil Environment
In year one, mean soil percent organic matter (% OM) was 9.88% prior to treatment application. Chemical analysis showed variability across treatments for pH and chemical composition (Table 1.2). Soil nutrient test results taken in year two indicated that there was a significant treatment effect on soil buffer pH (F = 3.79; df = 4, 29; P = 0.0153), % OM (F = 7.19; df = 4, 29; P =
0.0005), soil calcium (F = 3.91; df = 4, 29; P = 0.0134), soil manganese (F = 2.48; df = 4, 29; P = 0.0694), and cation exchange capacity (CEC) (F = 6.13; df = 4, 29; P = 0.0014) (Table 1.3). Buffer pH was significantly greater in bark mulch and shaving treatments compared to the control treatment. The sawdust treatment had significantly greater mean % OM than shaving, bark, and wood chip treatments and significantly greater soil calcium, soil manganese, and CEC compared to the wood chip treatment. The control treatment also had significantly greater % OM and CEC compared to the wood chip treatment.
Table 1.3. Soil analysis treatment means from soil sampling after harvest. Results include soil pH and buffer pH, percent organic matter (% OM), all major nutrients (nitrate ($\text{NO}_3$), ammonium ($\text{NH}_4$), phosphorus (P), potassium (K), and calcium (Ca)), micronutrients with statistical significance (manganese (Mn)), and cation exchange capacity (CEC).

<table>
<thead>
<tr>
<th>Treatments</th>
<th>pH</th>
<th>buffer pH</th>
<th>% OM</th>
<th>$\text{NO}_3$ (mg/kg)</th>
<th>$\text{NH}_4$ (mg/kg)</th>
<th>P (kg/ha)</th>
<th>K (kg/ha)</th>
<th>Mg (kg/ha)</th>
<th>Ca (kg/ha)</th>
<th>Mn (mg/kg)</th>
<th>CEC (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sawdust</td>
<td>5.1</td>
<td>5.62 ab</td>
<td>9.95 a</td>
<td>0.0</td>
<td>1.67</td>
<td>3.47</td>
<td>160.10</td>
<td>100.32</td>
<td>570.13 a</td>
<td>9.90 a</td>
<td>3.63 a</td>
</tr>
<tr>
<td>Shavings</td>
<td>5.1</td>
<td>5.67 a</td>
<td>8.33 bc</td>
<td>0.0</td>
<td>2.17</td>
<td>3.22</td>
<td>150.40</td>
<td>81.07</td>
<td>437.70 ab</td>
<td>7.60 ab</td>
<td>3.12 abc</td>
</tr>
<tr>
<td>Bark</td>
<td>5.2</td>
<td>5.67 a</td>
<td>7.70 bc</td>
<td>0.0</td>
<td>2.33</td>
<td>3.13</td>
<td>146.83</td>
<td>93.77</td>
<td>428.33 ab</td>
<td>7.32 ab</td>
<td>3.08 bc</td>
</tr>
<tr>
<td>Chips</td>
<td>5.2</td>
<td>5.65 ab</td>
<td>7.45 c</td>
<td>0.0</td>
<td>1.33</td>
<td>3.15</td>
<td>147.20</td>
<td>66.33</td>
<td>362.40 b</td>
<td>6.43 b</td>
<td>2.92 c</td>
</tr>
<tr>
<td>Control</td>
<td>5.2</td>
<td>5.57 b</td>
<td>9.08 ab</td>
<td>0.17</td>
<td>1.67</td>
<td>3.32</td>
<td>160.10</td>
<td>87.82</td>
<td>536.72 ab</td>
<td>7.77 ab</td>
<td>3.58 ab</td>
</tr>
</tbody>
</table>
There was a significant effect of treatment (P < 0.0001) and date (P < 0.0001) on soil moisture during both years of the study (Figure 1.1). There was also a significant interaction of treatment by date in 2020 (P < 0.0001). No treatment had significantly greater mean soil moisture compared to the control, except for bark mulch on June 16, 2020 (bark mean = 25.75% VWC, control mean = 22.78% VWC, P = 0.0138). Mean soil moisture for the shaving treatment was significantly greater than the wood chip treatment during 50% of the 2020 sampling dates. Overall mean soil moisture was significantly greater in the shaving and control treatments compared to the wood chip treatment during both years. Soil moisture ranged from 2.8% to 46.6% in 2020 and from 10.4% to 45.3% in 2021. No significant treatment effects on soil temperature were detected throughout the 2020 and 2021 field seasons. Only date had a significant effect on soil temperature (P < 0.0001) during both years of the study. Soil temperature ranged from 22.2°C to 37.6°C in 2020 and from 16.4°C to 43.7°C in 2021.
Figure 1.1. Mean soil moisture content by date and mulch type for wild blueberry prune (2020) and crop (2021) years. The blue line represents total rainfall and melted snow amounts. Error bars represent the SEM (n = 240). Letters indicate pairwise differences within each graph. Graph letters from top down correspond to legend treatments top down. Precipitation data is from NOAA National Environmental Satellite, Data, and Information Service Record of Climatological Observations for Prospect, ME weather station.
1.4.2 Plant Growth

1.4.2.1 Prune Year Measures

There was not a significant treatment effect on stem density measured at the end of the prune year (F = 0.51; df = 4, 59; P = 0.7273). There was a significant treatment effect on the change in stem density from the beginning to the end of the prune year (F = 4.97; df = 4, 59; P = 0.0017). The change in stem density for sawdust and shaving treatments was significantly more than that of the control treatment. There was a trend toward increased stem density for all mulch treatments (sawdust by 23.93%, shavings by 34.07%, bark by 11.47%, chips by 18.74%) observed from June to August 2020. Control plots decreased in stem density by 9.47% in the same time span.

1.4.2.2 Crop Year Measures

There was a significant treatment effect on both stem height (F = 4.82; df = 4, 299; P = 0.0009) and number of buds per stem (F = 4.04; df = 4, 299; P = 0.0033). Stems were significantly taller in sawdust and shaving treatments compared to the control (Figure 1.2). Mean number of buds per stem was also significantly greater in all mulch treatments compared to the control.

Figure 1.2. Mean stem height and number of buds per stem by mulch type. Error bars represent the SEM (n = 300). Letters indicate pairwise differences.
There was a significant treatment effect on the number of flowers per stem ($F = 3.81; \text{df} = 4, 299; P = 0.0049$), blue fruit per stem ($F = 2.99; \text{df} = 4, 299; P = 0.0191$), and yield ($F = 3.66; \text{df} = 4, 29; P = 0.0176$). The sawdust treatment had significantly more flowers per stem compared to the control treatment during bloom 2021 (Table 1.4). The bark mulch treatment had significantly more blue fruit per stem compared to the control treatment by the final date of fruit counts during harvest. Shaving and sawdust treatments had the greatest mean total yield. No significant differences were detected among treatments for the number of green fruit per stem, 100-berry weights or sugar content as indicated by Brix measurements.

Table 1.4. Blueberry flower and fruit counts, plot yield, berry sample weights and Brix sugar content as affected by mulch treatments during wild blueberry crop year (2021) growing season.

<table>
<thead>
<tr>
<th>Treatments</th>
<th># Flowers per stem</th>
<th># Green fruit per stem</th>
<th># Blue fruit per stem</th>
<th>Yield (kg/ha)</th>
<th>100-berry weights (g)</th>
<th>°Brix</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sawdust</td>
<td>13.20 a</td>
<td>7.38</td>
<td>4.07 ab</td>
<td>2516 a</td>
<td>34.00</td>
<td>10.79</td>
</tr>
<tr>
<td>Shavings</td>
<td>10.32 ab</td>
<td>6.43</td>
<td>4.57 ab</td>
<td>2646 a</td>
<td>32.00</td>
<td>10.89</td>
</tr>
<tr>
<td>Bark</td>
<td>11.85 ab</td>
<td>7.33</td>
<td>5.57 a</td>
<td>2168 ab</td>
<td>31.00</td>
<td>10.80</td>
</tr>
<tr>
<td>Chips</td>
<td>11.37 ab</td>
<td>7.17</td>
<td>4.65 ab</td>
<td>2077 ab</td>
<td>34.17</td>
<td>11.11</td>
</tr>
<tr>
<td>Control</td>
<td>8.58 b</td>
<td>5.58</td>
<td>3.52 b</td>
<td>1680 b</td>
<td>32.33</td>
<td>11.28</td>
</tr>
</tbody>
</table>

1.4.2.3 Both Year Measures

There was a significant effect of treatment, date, and the interaction of treatment by date for blueberry cover during 2020. Control plots had significantly greater percent blueberry cover than all mulch treatments in June 2020. By July and August, there were no statistically significant differences in blueberry cover among treatments. All mulch treatments increased in blueberry cover by August 2020 while blueberry cover in the control treatment decreased. This trend was also reflected in treatment changes in stem density during the prune year. There were no significant differences detected in blueberry cover in 2021.
Table 1.5. Blueberry pest pressure as affected by mulch treatments during wild blueberry prune (2020) and crop (2021) years.

<table>
<thead>
<tr>
<th>Year and Treatment</th>
<th>Weed Density (plants/m²)</th>
<th>Weed Cover (%)</th>
<th>Disease Density (stems/m²)</th>
<th>Disease Cover (%)</th>
<th>Insect Density (stems/m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2020</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sawdust</td>
<td>101.95</td>
<td>29.78</td>
<td>58.71 b</td>
<td>8.24 b</td>
<td>14.94</td>
</tr>
<tr>
<td>Shavings</td>
<td>133.41</td>
<td>27.92</td>
<td>52.85 b</td>
<td>5.00 b</td>
<td>14.49</td>
</tr>
<tr>
<td>Bark</td>
<td>112.09</td>
<td>26.96</td>
<td>64.79 b</td>
<td>6.83 b</td>
<td>11.94</td>
</tr>
<tr>
<td>Chips</td>
<td>126.65</td>
<td>27.88</td>
<td>70.80 b</td>
<td>7.08 b</td>
<td>11.79</td>
</tr>
<tr>
<td>Control</td>
<td>97.00</td>
<td>24.21</td>
<td>156.76 a</td>
<td>24.14 a</td>
<td>15.47</td>
</tr>
<tr>
<td>2021</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sawdust</td>
<td>74.21</td>
<td>25.94</td>
<td>78.38</td>
<td>12.94</td>
<td>7.21</td>
</tr>
<tr>
<td>Shavings</td>
<td>76.46</td>
<td>21.79</td>
<td>92.57</td>
<td>15.71</td>
<td>14.75</td>
</tr>
<tr>
<td>Bark</td>
<td>76.58</td>
<td>15.69</td>
<td>96.17</td>
<td>16.65</td>
<td>7.77</td>
</tr>
<tr>
<td>Chips</td>
<td>80.41</td>
<td>21.06</td>
<td>81.87</td>
<td>15.35</td>
<td>9.23</td>
</tr>
<tr>
<td>Control</td>
<td>70.95</td>
<td>19.02</td>
<td>112.50</td>
<td>19.42</td>
<td>10.14</td>
</tr>
</tbody>
</table>

Year and Model Effect

<table>
<thead>
<tr>
<th>Mixed Model Effect p-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Block (B)</td>
</tr>
<tr>
<td>Treatment (T)</td>
</tr>
<tr>
<td>Date (D)</td>
</tr>
<tr>
<td>T x D</td>
</tr>
<tr>
<td>2021 Block</td>
</tr>
<tr>
<td>Treatment (T)</td>
</tr>
<tr>
<td>Date (D)</td>
</tr>
<tr>
<td>T x D</td>
</tr>
</tbody>
</table>
Because insect damage did not vary in percent cover, only insect density data is presented to show the number of stems where insect pests were present (Table 1.5). In 2020, there was no significant treatment effect on weed density, weed cover, or insect density. There was a significant effect of date on weed density, weed cover, and insect density during 2020. These measures peaked in July, the middle of the growing season. The most abundant broadleaf weeds observed in 2020 were common cinquefoil (*Potentilla simplex* Michaux.), goldenrod (*Solidago* spp.), and golden clover (*Trifolium aureum* Pollich.). Grasses also made up a large fraction of weed plants observed. The most abundant insects observed were blueberry gall midge (*Dasineura oxycoccana* Johnson), red-striped fireworm (*Aroga trialbamaculella* Chambers), and flea beetle (*Altica sylvia* Malloch). There were no significant interactions for weeds or insects.

There was a significant effect of treatment, date, and the interaction of treatment by date on disease density and cover in 2020. No significant differences were detected in June 2020. In July and August 2020, the control treatment had significantly greater disease density and cover compared to all mulch treatments. In July, the control had 141%, 157%, 141%, and 129% more disease cover than sawdust, shaving, bark, and wood chip treatments, respectively. In August, control plots had 75%, 118%, 91%, and 91% more disease cover than sawdust, shaving, bark, and wood chip treatments, respectively. The most abundant diseases observed were leaf spot diseases, primarily *Sphaerulina* leaf spot caused by *Sphaerulina vacinii*, which accounted for 99.7% of disease observations per stem and therefore accounted for significant differences among treatments.

In 2021, there were no significant treatment effects on density or cover for weeds, insects, and diseases. There was a significant effect of date on weed density. The total number of weeds significantly increased from June to July 2021. The most abundant broadleaf weeds
observed in 2021 were common cinquefoil (*Potentilla simplex* Michaux.), goldenrod (*Solidago* spp.), and common vetch (*Vicia sativa* L.). Grasses again made up a large fraction of weed plants observed. The most abundant insects observed were blueberry gall midge (*Dasineura oxycoecana* Johnson) and blueberry thrips (*Frankliniella vaccinii* Morgan and *Catinathrips kainos* O’Neill). There was a significant effect of date on disease density and cover in 2021, which significantly increased from June to July 2021 across all plots. The most abundant diseases observed were again leaf spot diseases, which accounted for 97.6% of disease observations per stem.

In 2020, there was a significant effect of treatment (*P* = 0.0001), date (*P* < 0.0001), and the interaction of treatment by date (*P* = 0.0004) on foliar chlorophyll content. Overall mean chlorophyll content in 2020 was significantly greater in sawdust (31.48 SPAD units), shaving (30.71 SPAD units), and wood chip (30.74 SPAD units) treatments compared to the control (29.48 SPAD units). No significant differences were detected in June 2020. Control plots were significantly lower in chlorophyll content than sawdust, shaving, and wood chip treatments during July 2020 and significantly lower than the wood chip treatment during August 2020 (Figure 1.3). In 2021, there was a significant effect of treatment (*P* < 0.0001) and date (*P* < 0.0001) on foliar chlorophyll content. Sawdust treatments had significantly greater overall mean chlorophyll content (26.19 SPAD units) compared to all other treatments in 2021.
Figure 1.3. Monthly mean leaf tissue chlorophyll content in SPAD units. Error bars represent the SEM (n = 1,200). Letters indicate pairwise differences. Graph letters from top down correspond to legend treatments left to right.

1.5 Discussion

Overall, mulching was beneficial to wild blueberry plant growth in both prune and crop years. All mulch treatments resulted in significantly fewer diseased stems with less disease cover in the first year and significantly more buds per stem by the second year compared to the control treatment. Sawdust and shaving treatments had significantly taller stems, a significant increase in stem density, and significantly greater yields compared to the control treatment. However, no mulch treatment demonstrated significantly greater overall mean soil moisture compared to the control, suggesting that the 1.27 cm mulch layer used in this study was not thick enough to conserve significantly more soil moisture than the control treatment. This thickness of mulch was chosen as an economically and physically feasible amount of material for growers to apply to
entire fields, yet further research should investigate different thicknesses of these mulches. Smaller particles sizes promoted overall blueberry plant growth and can be a useful tool for growers to increase yields more effectively than larger particle sizes such as wood chips.

1.5.1 Soil Environment

Our soil analysis showed soil organic matter, calcium, manganese, and CEC were significantly greater in the sawdust treatment, the finest particle size, than the wood chip treatment, the largest particle size. These differences may have been impacted by sampling error if mulch particles, along with blueberry roots and leaves, were not completely removed from soil cores during sampling. Smaller particles of sawdust were especially difficult to remove from samples compared to larger wood chips if they mixed into soil cores during sampling. Decomposition rate may have also impacted these results. Duryea et al. (1999) reported that wood mulches such as pine bark (*Pinus elliottii* and *P. taeda*) showed 3% to 7% decomposition after one year. However, Allison (1965) showed that decomposition rates were faster for finer shortleaf pine (*Pinus echinata*) wood particles. The sawdust in this experiment may have decomposed slightly faster than the larger particle size treatments, increasing organic matter and therefore CEC. In an analysis of 347 A horizons and 696 B horizons of New Zealand soils, Parfitt et al. (1995) showed that most CEC arose from soil organic matter compared to clay content and other clay minerals. Organic matter also plays an important role in soil buffer pH for acid soils (Helling et al. 1964; Keeney and Corey, 1963; Magdoff and Bartlett, 1985). Generally, higher organic matter content is correlated with higher buffer capacity. Because the results of the soil analysis may be influenced by the mulches mixing with the soil during sampling and the difficulty of separating finer particles from samples, future research should follow mulch materials for more than one production cycle to record the effects of their decomposition on the soil environment.
The control treatment performed similarly or better than mulch treatments regarding soil moisture during both years (Figure 1.1). The instability and subjection to wind and erosion may have contributed to poor performance of the sawdust treatment in retaining soil moisture (DeGomez and Smagula, 1990ab). Other particle size studies found similar results regarding insignificance among treatments. Diaz et al. (2005) found that a 5 cm application of tephra mulch resulted in no significant differences among particle size for soil water evaporation. Gilman and Grabosky (2004) found that pine bark mulch thickness and particle size did not significantly affect stem xylem water potential or trunk diameter in newly planted live oaks (Quercus virginiana Mill.). However, some studies have found that finer particle sizes were more effective than larger particles in preserving soil temperature and reducing soil water evaporation (Xie et al. 2010; Qui et al. 2014).

Soil temperature varied by date throughout each season, as expected, but there were no significant differences among treatments. This suggests that particle size, at the thickness applied in this study (1.27 cm), did not affect soil temperature. A thicker application may have improved soil moisture retention and shown differences in soil temperature. Pakdel et al. (2011) found significant reductions in soil temperature and increases in soil moisture with sawdust, wood chip, and gravel mulch at thicknesses of 5 cm to 15 cm. Van Donk (2011) found that addition of wood chip mulch at 2.5, 5, and 10 cm thicknesses increased soil water content with the most significant differences measured at the 5 and 10 cm thicknesses. Economically, a shallower application may be required for whole-field mulching, yet a thicker application may be used to increase soil moisture in lower-yielding fields with greater water deficits. When they do apply mulch, Maine growers primarily apply pine wood chips. However, because this particle size retained the least soil moisture, growers should consider using a finer particle size when applying mulch.
Soil moisture in this study increased following precipitation events, especially those that accumulated more than one cm of rain, which occurred during June through August in 2020 and May through July in 2021 (Figure 1.1). Future research should measure soil moisture at more frequent intervals to determine how long after rain events the ground stays moist under each treatment.

### 1.5.2 Plant Growth

Mulch can improve wild blueberry growth, especially through the spread of rhizomes (Kender and Eggert, 1966; Yarborough, 2012). This spread contributes to new stem growth, which is supported by our stem density and blueberry cover results. Although control treatments started with significantly more blueberry cover in June 2020, only mulch treatments had a positive change in stem density by the end of the prune year.

Mulch reduces diseases such as leaf spot and mummy berry by covering infected tissues, reducing inoculum, and preventing dispersal of overwintering spores (Alfieri, 1991; Drummond et al. 2009). The results of this study support these findings. All mulch treatments significantly reduced disease relative to the control in the first year. By reducing disease pressure, mulch can mitigate plant stress, likely influencing the positive responses seen in stem density and the number of buds per stem for all mulches, as well as stem height and yield for sawdust and shaving treatments compared to the control treatment. These results agree with previous research that showed leaf spot incidence in wild blueberry fields was inversely related to flower bud set and berry production (Annis and Stubbs, 2004; Ojambo et al. 2002). A trend toward greater leaf spot disease cover in control plots during both years and fewer mean number of buds, flowers, and blue fruit per stem in year two was observed. Our results also support the typical phenology of common weed, insect, and disease pests in wild blueberry (Yarborough et al. 2009). While the
most weed growth and emergence of adult insects peaked mid-season, disease infection significantly increased each month until the end of the growing season.

Nitrogen immobilization or “locking” has been suggested as a concern when applying mulches to crops. Sønsteby et al. (2004) showed that bark mulch slightly reduced foliar nitrogen and significantly reduced soil nitrate and ammonium levels in strawberry. Krewer et al. (2008) suggested pine bark mulch tied up more applied nitrogen than pine straw in rabbiteye blueberry. As soil microbes use nitrogen to break down the applied organic matter, a shallow depletion zone may form at the interface of the soil surface and mulch layer. Our results showed that control plots had the lowest overall mean chlorophyll content in year one and were among the lowest in year two (Figure 1.3). This indicates the mulch application resulted in greater nitrogen in plant tissues and that nitrogen locking was not observed in mulch treatments. These results agree with other studies that have shown wood mulches to increase nitrogen levels in soil and foliage (Arthur and Wang, 1999; Cregg and Schutzki, 2009).

1.5.3 Implications for Farmers

Wild blueberry farmers face many climate challenges like erratic frost and drought events (Fernandez et al. 2020). Summer 2020 in Maine was the most severe short-term drought since the early 2000s and the worst fire season in the last decade (Birkel, 2020a). The summer of 2021 was also similar in its temperature anomalies. In temperate regions like Maine, irrigation usage may range from “not at all” to “frequent” due to uneven rainfall and unpredictable weather patterns, making it difficult to predict irrigation demand and total cost from year to year (Dalton and Yarborough, 2004). Given the highly variable rainfall patterns across the wild blueberry production regions of Maine, irrigation may help increase yields when rainfall is limited, but poses a financial risk to farmers when rainfall events are largely sporadic. Cultural management
options such as mulching can in turn be a relatively easy way to improve wild blueberry production.

Growers should consider using a thicker application and focus this application toward lower-yielding portions of fields. A thicker application will increase the amount of material needed and therefore cost for material but can improve overall productivity. Applications should not exceed ten centimeters (DeGomez and Smagula, 1990b). The standard practice used by the Natural Resources Conservation Service is a 5.08 cm application (NRCS, 2017). Growers might consider top-dressing finer particles with a larger particle size to weigh down the lightweight material, increase stability, and slow decomposition rate to reduce the need for reapplication. Applying a second material would be more expensive, however, the finest particle size treatments were the least expensive materials to purchase for this study (Table 1.2). Wood chips were the most expensive material purchased. Costs may vary depending on the source, especially if growers can produce their own mulch on-site. Future research should investigate the effectiveness of different combinations of thicknesses and particle sizes in wild blueberry.

Research on the comparative effects of mulching with materials that have a high decomposition rate to increase soil organic matter versus materials with a slow decomposition rate to maintain a top layer would also be valuable.

If interested in using mulch as a crop management tool, growers should consider using finer particle sizes rather than wood chips. Over time, however, larger particle size mulches may perform better than finer particle size mulches if the finer sizes are not reapplied, due to their tendency to erode (DeGomez and Smagula, 1990b). From this study, any softwood mulch particle size will improve plant health.
CHAPTER 2: FERTILITY EFFECTS ON BLUEBERRY GALL MIDGE (DASINEURA OXYCOCCANA (JOHNSON) (DIPTERA: CECIDOMYIIDAE)) IN WILD BLUEBERRY (VACCINIUM ANGUSTIFOLIUM AITON.)

2.1 Chapter Abstract

Blueberry gall midge, Dasineura oxycoccana (Johnson) (Diptera: Cecidomyiidae) is a pest in American cranberry, Vaccinium macrocarpon Aiton (Ericales: Ericaceae) and wild blueberry, Vaccinium angustifolium Aiton (Ericales: Ericaceae), and has been observed in areas of high soil and foliar nutrient levels. New management strategies, including fertilization, will need to be altered to sustain wild blueberry production under climate change and, in turn, may impact the occurrence of this pest. The purpose of this study was to measure the effect of diammonium phosphate fertilizer application on gall density and the combined effects of fertilizer application with and without galling on wild blueberry systems. This study was conducted at two field sites in Jonesboro and Washington, ME during 2020 and 2021. Fertilizer application resulted in significantly greater gall density during the prune year in Jonesboro and both years in Washington. Foliar percent nitrogen and phosphorus had a significant positive linear relationship with gall density. Galling without fertilizer application resulted in significantly shorter stems with fewer buds per stem at both sites compared to fertilized stems without galls. Fertilizer application without galling resulted in the greatest number of buds, flowers, and berries per stem at the Washington site. Mean stem height and total yield at this site were greater in fertilizer treatments even when galling was present. Growers applying fertilizers should monitor blueberry gall midge field infestation levels due to our findings that DAP fertilizer impacted the economic thresholds for this pest and DAP fertilizer with galling impacted wild blueberry development and yield.
2.2 Introduction

2.2.1 Wild Blueberry Systems

Wild blueberry, *Vaccinium angustifolium* Aiton (Ericales: Ericaceae), is a perennial fruit crop managed in large expansions of low-lying shrubs on a two-year production cycle. Growers either mow or burn fields in the first year of the cycle, the “prune” year, to stimulate vegetative growth. Plants develop flowers and bear fruit in the second “crop” year, when yields are harvested. Maine produces 10% of all blueberries in North America, making it a significant blueberry producer worldwide (Yarborough, 2015). Wild blueberries are an iconic economic and cultural fruit crop in the state of Maine, which supports more than 450 growers. In 2020, wild blueberry growers produced 21.5 million kilograms of wild blueberries harvested from 8,377 hectares for a price of $1.81/kg of fresh berries and $1.32/kg of processed berries (USDA NASS, 2021b). Compared to the previous five-year-average (2015-2019), this was a 1.67% increase in area harvested and 58% increase in price per kilogram of processed berries, but a 41% decrease in kilograms per hectare produced and 43% decrease in price per kilogram of fresh berries.

Climate change has already had a significant impact on wild blueberry systems, especially through increasing rates of warming temperatures. The Northeast is warming faster than the rest of the United States (Fernandez et al. 2020) with the most significant temperature increases occurring in the Downeast region of Maine, where most wild blueberries are grown in the state (Tasnim et al. 2021). Rising temperatures can significantly impact wild blueberry growth and development, resulting in major changes to the wild blueberry nutrient economy and therefore grower fertilization regimes (Tasnim et al. 2020; 2021). These changes may in turn have significant impacts on pests responding to the wild blueberry nutrient environment.
2.2.2 Blueberry Gall Midge

Blueberry gall midge, *Dasineura oxycoccana* (Johnson) (Diptera: Cecidomyiidae), is an emerging insect pest that was discovered in Maine wild blueberry fields in 2003 (Collins and Drummond, 2019). It has been deemed the most important insect pest of blueberries grown in the southern United States (Dernisky et al. 2005) and has since spread northward, steadily increasing in occurrence and density across wild blueberry fields in Maine (Collins and Drummond, 2019).

The blueberry gall midge life cycle specific to Maine wild blueberry systems was described by Collins and Drummond (2017). They described adults as small, mosquito-like flies, less than two millimeters long, which makes them difficult to see with the naked eye. Females deposit eggs in the terminal shoots of new blueberry growth. Larvae develop through three instars while feeding on new growth. This feeding causes leaves to dimple and curl, forming leaf galls on the tips of stems. Blueberry gall midge is found in greater abundance in prune than crop fields, most likely due to the requirement of actively developing plant tissues for gall formation by many gall-inducing insects (Rohfritsch, 1992). The prune year in wild blueberry is the vegetative year in which new vegetative shoots grow from rhizomes (Yarborough and Smagula, 2015), thus providing said attractive tissues for this pest.

Galling can reduce the number of buds and flowers per wild blueberry stem and therefore total fruit set, which can cause yield loss up to 50% for any single infested stem (Collins and Drummond, 2019; Yarborough et al. 2017). Plant response to gall midge damage can result in excessive branching in the prune year, thus delaying flower bud development, increasing susceptibility to winterkill and disease, and decreasing the number of viable flower buds and therefore yields in the crop year (Collins and Drummond, 2017; Cutler and Sproule, 2011).
Increased fertilization is believed to increase pest presence by providing more vegetative growth and therefore habitat for the insects (Reekie, MacKenzie, and Lees, 2009). Yarborough et al. (2017) suggested that leaf nutrition influences gall midge levels based on observations of higher gall midge abundance in areas of increasing leaf boron and phosphorus levels. This pest is already known to respond to excess nitrogen in cranberry, with a preference to lay eggs on newer tissues (Leduc and Turcotte, 2004). Phosphorus is also often a limiting factor for many organisms competing for available nutrients in the environment (Drinkwater and Snapp, 2007; Faucon et al. 2015). Diammonium phosphate (DAP [18-46-0]) is the standard conventional fertilizer used by wild blueberry growers with the goal of adding nitrogen and phosphorus to the crop for better yields. Increasing nutrient inputs, primarily through nitrogen and phosphorus levels, may therefore increase gall midge infestations in Maine wild blueberry fields, but more research is needed to validate these hypotheses. Growers sometimes forego fertilization when market prices are low and budgets require management to be spent elsewhere, but this may change with climate change. It is valuable to know whether adding specific nutrients to this crop attracts blueberry gall midge and to what extent damage is caused if it does. This will allow growers to determine whether they want to budget finances for fertilizer costs or to put resources towards other expenses. This study aims to gain more insight on how nutrient management could be used to manage gall midge activity in wild blueberry systems as part of the wild blueberry integrated pest management program.

The objectives of this study were to determine (1) the effect of DAP application on plant growth, soil nutrients, and gall density, (2) the effect of galling on plant growth and berry development as indicated by plant height and the number of buds, flowers, and berries per stem, (3) the combined effects of DAP application with galling on wild blueberry and their
implications for pest management. We hypothesized that gall density would increase where there were high levels of nitrogen and phosphorus leaf nutrient content. We also hypothesized that the addition of DAP where gall midge was present would increase gall midge damage and therefore crop loss due to increased gall density.

2.3 Materials and Methods

2.3.1 Study Sites
This study was located at two conventionally managed fields in Jonesboro and Washington, Maine. The Jonesboro site was located at the University of Maine Blueberry Hill Farm Experiment Station, the only university-based wild blueberry research station in the United States. This field was in the crop year in 2020 and prune year in 2021. The Washington site was in the prune year in 2020 and crop year in 2021. Both sites had blueberry gall midge infestations detected in 2019. The experiment was conducted from May 2020 to September 2021 to cover one full production cycle at both sites. The experiment was a randomized complete block split-plot design with eight replicates of each treatment. Each plot was 1.83 m by 9.14 (16.72 m²) with 0.91 m of wild blueberry between plots to serve as a buffer.

2.3.2 Treatments
The whole plot factor was blueberry gall level, either present or absent. A “present” gall treatment only had stems with galls flagged for repeated measures. An “absent” gall treatment only had stems without any evidence of galls flagged and was monitored throughout the season to ensure that those without leaf galls were not later infested before further measurements were taken. New stems were flagged if the original stem developed a gall. The split plot factor was fertility level, either fertilized with DAP or unfertilized (no DAP applied). Stems without galls or fertilizer applied acted as a control treatment. Fertilized plots in both locations had 493.18 kg/ha
DAP (approx. 0.45 kg/split-plot) applied on June 10, 2020. This is the highest recommended rate for plants with leaf nitrogen levels below 1.76% and leaf phosphorus levels below 0.111%. Application rates were based on low nitrogen and phosphorus levels indicated in a 2018 plant tissue report at the Jonesboro site and UMaine Extension recommendations (Yarborough and Smagula, 2013).

2.3.3 Data Collection

2.3.3.1 Soil & Leaf Samples

Soil samples were taken at the end of the study with a hand-held soil probe to a depth of 10 cm from each plot in Jonesboro on August 10, 2021 and Washington on July 27, 2021. Samples were submitted to the Maine Agricultural and Forest Experiment Station Analytical Laboratory for standard field soil tests (Hoskins, 1997).

Leaf samples were taken from each plot on July 7, 2021 in Jonesboro and on July 6, 2021 in Washington. Leaf samples were taken by stripping all leaves from 30 stems/plot. Stems that had leaves stripped corresponded to the same plot treatment combinations but were separate from flagged stems. Samples were brought to the University of Maine Soil Testing Service in Orono, ME for standard foliar analysis. Plant material was dried, ground, and analyzed for nitrogen using a Leco Tru-Mac C/N Analyzer. Other nutrients were analyzed after low temperature dry ashing and ash dissolution by inductively coupled plasma optical emission spectrometry (Kalra and Maynard, 1991).

2.3.3.2 Gall Midge

Gall density was measured at both field sites for both crop and prune years by counting the number of stems with galls in two 0.37-m² quadrats per plot. One permanent quadrat was placed in each split-plot section. Only recently formed galls, indicated by fully closed leaf galls with no
signs of opening or uncurling of the leaves, were counted. Gall density was recorded at the Jonesboro site in 2020 on June 17; July 2, 7, 22, 28; and August 12. It was recorded in 2021 on June 2, 9, 16, 25, 30; July 7, 14, 21; and August 3. Gall density was recorded at the Washington site in 2020 on June 16; July 1, 10, 23, 30; August 21; and September 4, 16. It was recorded in 2021 on June 1, 10, 17, 24; and July 1, 6, 13, 19, 27. Recording gall density was stopped when no new galls were detected in plot quadrats during a prune year or when a field was harvested during a crop year.

2.3.3.3 Plant Growth

2.3.3.3.1 Prune Year Measures

Stem height and bud number were measured for each flagged stem in each plot at both sites. Stem density was measured by counting the number of stems in each quadrat at the beginning of the experiment and end of the prune year at both sites. Beginning stem density measurements were recorded on June 16 and 17, 2020 in Washington and Jonesboro, respectively. Jonesboro stem height and end of prune year stem density were measured on August 3, 2021 and the number of buds per stem was measured on August 24, 2021. Stem height, bud number, and end of prune year stem density were measured in Washington on September 4, 2020. Percent infestation was calculated by dividing gall density by stem density. Percent infestation calculations only used gall densities from the same dates stem density was recorded.

2.3.3.3.2 Crop Year Measures

Berry count and weight data for each flagged stem were collected during the crop year at each field site. For count data, the number of remaining green, red, blue, and diseased/over-ripe berries per stem were counted. Only healthy blueberries were considered marketable, hand-picked, and weighed for berry weight per stem. Berry weight per stem was used to extrapolate
potential yield per hectare based on an approximate stem density of 500 stems/m². Berries in each quadrat were hand raked and weighed. These measurements were taken on August 12 and 13, 2020 in Jonesboro and on July 27, 2021, in Washington.

The Jonesboro field site experienced drought and severe leaf spot disease resulting in significant defoliation during harvest 2020. Therefore, harvest data for this site is not presented. Flower, green fruit, and red fruit counts were added to data collection for the Washington field site in 2021 to account for yield loss in Jonesboro 2020. The number of open flowers per stem was recorded in Washington on May 21, 2021. The numbers of green, red, and blue fruit per stem were recorded in Washington before and during fruit ripening on June 17 and July 6, 2021, respectively. Because the Jonesboro field was in the crop year when the experiment was started in 2020, new stems were flagged for prune year measurements in 2021. To allow the Jonesboro field site to be mowed between its crop year 2020 and prune year 2021, stakes delineating plots at this site were removed and replaced with metal washers. A metal detector was used to re-stake plots at the beginning of the 2021 field season.

2.3.3.3 Both Year Measures

Blueberry cover was measured during both years on the same dates as gall density in each quadrat using a 0-6 ranking system with even intervals that sum to 100%, where: 0 = not present, 1 = ≤1%-16.67%, 2 = >16.67%-33.33%, 3 = >33.33%-50%, 4 = >50%-66.67%, 5 = >66.67%-83.33% and 6 = >83.33%-100%. Leaf chlorophyll content was measured as an indicator of nitrogen (Xiong et al. 2015) monthly during the summer using a SPAD Chlorophyll Meter (SPAD 502; Minolta Corp., Osaka, Japan). One measurement was taken from each of two leaves per flagged stem in each plot, one on the lowermost and one on the uppermost portions of the stem. SPAD measurements were recorded at the Jonesboro site in 2020 on June 17, July 9, and
August 12 and in 2021 on June 16 and July 23. SPAD measurements were recorded at the Washington site in 2020 on June 18, July 10, and August 21 and in 2021 on June 17 and July 19.

2.3.3.4 End of Data Collection

Crop development during the 2021 season was at least two weeks ahead of development in 2020, which was also several weeks ahead of schedule compared to previous years. Because of this, berries were harvested much earlier in the season. August 2021 measurements were omitted after harvest.

2.3.4 Data Analysis

The effects of DAP application on gall midge density and soil environmental conditions, as well as the effects of gall midge infestation on plant growth (chlorophyll content, foliar nutrients, blueberry cover, stem density, stem height, bud development, yield) were statistically analyzed using JMP software (JMP® Pro, Version 15.2.0, SAS Institute, 2020b). One-way analysis of variance and two sample t-tests were used to compare treatments for data collected only once throughout the study, including foliar nutrients, end of prune year stem density, stem height, bud number per stem, flowers per stem, berries per stem, and soil nutrients. A t-test was also used for percent infestation analysis. Data that did not follow a normal response distribution were analyzed through randomization tests to confirm the results of analyses of variance that did not meet standard assumptions of normality or equal variances.

Repeated-measures analyses of gall density, blueberry cover, and chlorophyll content were done using a generalized linear mixed model (GLMM) to determine differences among treatments and their interaction with date throughout each growing season (SAS Institute, 2020a). In the GLMM, count data were modeled with a Poisson distribution and log link. Proportion data (ranks) were modeled with a binomial distribution and logit link. When
modeling rank data, each rank measurement was converted to the percent midpoint of the range that each rank represents. For each model’s effects, ‘block’ was used as a random effect and all other variables were considered fixed. For analysis of gall density and blueberry cover, fixed effects included date and fertility level. For analysis of chlorophyll content, fixed effects included date, gall level, and fertility level. All possible fixed-by-fixed effect interactions were included in all models.

Linear regression was used to model foliar percent nitrogen and phosphorus as continuous independent variables with mean gall density (number of galls/stem/m$^2$) as the dependent variable. This was used to test the hypothesis that gall density increases with increasing foliar nutrient levels. These two nutrients were chosen since they are the major nutrients in the fertilizer used in this study and because foliar nutrient levels are the primary method used to determine fertilizer needs in wild blueberry (Yarborough and Smagula, 2017) as opposed to soil nutrient levels. Both Jonesboro and Washington sites were pooled to determine the mean gall density. Only data from 2021 was used to perform the regression, the same year the foliar tests were taken.

Treatment means were separated by Tukey’s Highly Significant Differences test at the 0.05 significance level. For analysis of soil nutrients, gall density, stem density, blueberry cover, and percent infestation, only fertilized (DAP applied) and unfertilized (no DAP applied) treatments were compared. For analysis of foliar nutrients, chlorophyll content, stem height, number of buds per stem, flowers per stem, berries per stem, and berry weight per stem, all treatment combinations for stems with or without fertilizer applied and with or without galls were compared.
2.4 Results

The summers of 2020 and 2021 were the third and fourth warmest summers, respectively, in Maine since 1895 (Birkel, 2020a; 2021). Both received less precipitation than the historical mean. The winter of 2020 was the sixth warmest in Maine since 1895 (Birkel, 2020b).

2.4.1 Soil and Leaf Samples

At the Washington site, soil nitrogen (NH\textsubscript{4}) (t = 2.24; df = 1, 31; P = 0.0329) and phosphorus (P) (t = 2.77; df = 1, 31; P = 0.0094) were significantly greater in plots where DAP was applied (mean NH\textsubscript{4} = 4.06 mg/kg, mean P = 21.34 kg/ha) than in plots where DAP was not applied (mean NH\textsubscript{4} = 3.38 mg/kg, mean P = 16.86 kg/ha). There was not a significant treatment effect of DAP application on soil nitrogen (t = 0.26; df = 1, 31; P = 0.7953) or phosphorus (t = 0.24; df = 1, 31; P = 0.8098) at the Jonesboro site in 2021. Mean soil nitrogen was 3.88 mg/kg where DAP was applied and 3.75 mg/kg where DAP was not applied. Mean soil phosphorus was 15.43 kg/ha where DAP was applied and 15.12 kg/ha where DAP was not applied.

Considering all treatment combinations for DAP application and galling, fertilized stems with galls had the greatest mean foliar percent nitrogen (1.36%) compared to all other treatment combinations (F = 10.17; df = 3, 15; P = 0.0013) at the Washington site. There were no significant differences detected among treatment combinations in foliar percent phosphorus at the Washington site (F = 1.01; df = 3, 15; P = 0.4212). There were no significant differences detected among treatment combinations in foliar nitrogen (F = 2.20; df = 3, 15; P = 0.1407) or phosphorus (F = 1.98; df = 3, 15; P = 0.1709) at the Jonesboro site. There was not a significant treatment effect of DAP application on foliar phosphorus or nitrogen at either site.
2.4.2 Gall Midge

In 2020, there was not a significant treatment effect of DAP application on gall density at the Jonesboro site (P = 0.8223) (Figure 2.1). In the GLMM, only date produced a significant effect (P < 0.0001). There was a significant treatment effect of DAP application on gall density at the Washington site (Figure 2.2). Gall density at this site was significantly greater in fertilized plots than in unfertilized plots. In the GLMM, both DAP treatment (P < 0.0001) and date (P < 0.0001) had a significant effect on gall density at the Washington site. There were 87.93% more galls in Washington across the 2020 season, on average, in plots where DAP was applied compared to those where it was not. There were no significant interactions between date and DAP treatment in 2020.

In 2021, gall density was significantly greater in fertilized plots than in unfertilized plots in both Jonesboro (P = 0.0332) and Washington (P < 0.0001) (Figures 2.1 and 2.2). Date also had a significant effect on gall density at the Jonesboro site (P < 0.0001). There were no significant interactions between date and DAP treatment in 2021. There were approximately 25.79% more galls at the Jonesboro site and 121.71% more galls at the Washington site across the 2021 season in fertilized plots than in unfertilized plots. The number of galled stems recorded ranged from 0 to 27 galled stems/m² at the Jonesboro site and 0 to 59.45 galled stems/m² at the Washington site. The maximum number of galled stems recorded during a prune year was 59.45 galled stems/m² at the Washington site. The maximum number of galled stems recorded during a crop year was 32.43 galled stems/m² at the Jonesboro site.
Figure 2.1. Mean gall density by date at the Jonesboro field site during the 2020 wild blueberry crop cycle and 2021 wild blueberry prune cycle. Error bars represent the SEM (n = 32). *Significant at the 0.05 level of significance.

Figure 2.2. Mean gall density by date at the Washington field site during the 2020 wild blueberry prune cycle and 2021 wild blueberry crop cycle. Error bars represent the SEM (n = 32). *Significant at the 0.05 level of significance.
The effects of foliar percent nitrogen and phosphorus on gall density were significant in the regression analysis (Figures 2.3 and 2.4). Only 18-27% of the variance in gall density was explained by each nutrient, suggesting that other variables result in gall density increases in wild blueberry. Still, with significance of the analysis we conclude that there was a significant trend showing the number of galled stems/m² increased with increasing foliar percent nitrogen ($F = 23.14; P < 0.0001; r^2 = 0.27$) and percent phosphorus ($F = 13.25; P = 0.0006; r^2 = 0.18$).

Figure 2.3. Gall density increase as it relates to percent foliar nitrogen. The solid line represents the least square regression.
Figure 2.4. Gall density increase as it relates to percent foliar phosphorus. The solid line represents the least square regression.
2.4.3 Plant Growth

2.4.3.1 Prune Year Measures

There was not a significant treatment effect of DAP application on stem density at the end of the prune year in Jonesboro (t = 1.95; df = 1, 31; P = 0.0610) or Washington (t = 0.1.45; df = 1, 31; p = 0.1576). DAP application also did not have a significant effect on mean percent infestation (t = .46; df = 1, 63; P = 0.6484). Mean percent infestation was 0.42% where DAP was applied and 0.35% where it was not applied across both sites. Percent infestation ranged between 0% and 2.7%.

There were significant treatment effects on stem height (Jonesboro: F = 6.88; df = 3, 319; P = 0.0002, Washington: F = 67.93; df = 3, 319; P < 0.0001) and bud number per stem (Jonesboro: F = 4.57; df = 3, 319; P = 0.0038, Washington: F = 11.68; df = 3, 319; P < 0.0001). In Jonesboro, stems without galls were 15.80% taller than stems with galls. In Washington, fertilized stems without galls had the greatest mean stem height and number of buds per stem (Figures 2.5 and 2.6). Unfertilized stems with galls had significantly shorter stems and fewer buds per stem compared to fertilized stems without galls at both sites.
Figure 2.5. Mean stem height by gall level and fertility level combinations at the Washington field site during the 2020 wild blueberry prune cycle. Error bars represent the SEM (n = 320). Letters indicate pairwise differences.

Figure 2.6. Mean number of buds per stem by gall level and fertility level combinations at the Washington field site during the 2020 wild blueberry prune cycle. Error bars represent the SEM (n = 320). Letters indicate pairwise differences.
2.4.3.2 Crop Year Measures

There were significant differences amongst treatment combinations for the number of flowers per stem ($F = 15.36; \text{df} = 3, 319; P < 0.0001$) at the Washington site. Fertilized stems without galls had the greatest mean number of flowers per stem (Figure 2.7).

Figure 2.7. Mean number of flowers per stem by gall level and fertility level combinations at the Washington field site during the 2021 wild blueberry crop cycle. Error bars represent the SEM (n = 320). Letters indicate pairwise differences.

Before ripening, there were also significant treatment effects on the number of green berries per stem ($F = 14.16; \text{df} = 3, 319; P < 0.0001$). Fertilized stems without galls had significantly more green fruit than all other treatment combinations. During ripening, there were also significant treatment effects on the number of green berries per stem ($F = 11.13; \text{df} = 3, 319; P < 0.0001$) and blue berries per stem ($F = 8.65; \text{df} = 3, 319; P < 0.0001$). Fertilized stems without galls had significantly more green berries per stem than all other treatment combinations and significantly
more blue berries per stem than treatments without DAP applied. At harvest, there were significant treatment effects on the number of green berries per stem ($t = -2.19; df = 1,319; P = 0.0291$), blue berries per stem ($F = 15.05; df = 3, 319; P < 0.0001$) and berry weight ($F = 15.19; df = 3, 319; P < 0.0001$) at the Washington site. Galled stems had significantly fewer green berries (63.53%) and fewer ripe berries (37.03%) at harvest. There was also significantly more blue fruit per fertilized stem compared to stems without DAP applied (59.98% more). Blue fruit weight per stem was significantly heavier by 71.54% for fertilized stems than unfertilized stems ($t = 6.17; df = 1, 319; P < 0.0001$). No significant differences were detected for the number of red berries per stem at any stage of development recorded.

There was a significant treatment effect on yield ($F = 15.19; df = 3, 319; P < 0.0001$). Fertilized stems without galls had the greatest yield across all treatments (Figure 2.8). All fertilized stems had significantly greater yields than stems without DAP applied, regardless of gall presence. Fertilized stems without galls had 31.41%, 82.71%, and 85.25% greater yield than fertilized stems with galls, galled stems without DAP applied, and stems without galls or DAP applied, respectively (Table 2.1).
Figure 2.8. Mean total yield by treatment at the Washington field site during the 2021 wild blueberry crop cycle. Error bars represent the SEM (n = 320). Letters indicate pairwise differences.

Table 2.1 Treatment comparison of yield gain or loss. A (+) before percent values indicates that treatments in the left column had a yield gain compared to the corresponding treatment in the top row. A (–) before percent values indicates that treatments in the left column had a yield loss compared to the corresponding treatment in the top row.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield Gain or Loss (%)</th>
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<tbody>
<tr>
<td></td>
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</tr>
<tr>
<td>Not galled - No DAP</td>
<td>-</td>
</tr>
<tr>
<td>Not galled + DAP</td>
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<tr>
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<td>-</td>
</tr>
<tr>
<td>Galled + DAP</td>
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2.4.3.3 Both Year Measures

In 2020, there was a significant effect of date (P < 0.0001), DAP application (P < 0.0001), and their interaction (P = 0.0089) on blueberry cover in Washington. Blueberry cover was significantly greater in fertilized plots at the Washington site from mid-July through August.
2020. DAP application did not have a significant effect on blueberry cover at the Jonesboro site. Only date had a significant effect on blueberry cover (P < 0.0001) in Jonesboro. Blueberry cover at this site greatly decreased by August 2020 due to leaf spot disease defoliation. In 2021, there was a significant effect of date (P < 0.0001) and DAP application (P < 0.0001) on blueberry cover at both sites, as well as the interaction of date by DAP application (P < 0.0001) at the Washington site. Blueberry cover was significantly greater in plots where DAP was applied throughout June 2021 in Jonesboro and only on July 19 and 27, 2021 in Washington. Significant differences were primarily detected during the prune year at both sites.

In 2020, there were significant model effects of date, gall level, fertility level, the interactions of gall level by date and fertility level by date on chlorophyll content at the Jonesboro site (Table 2.2). At the Washington site, there were significant model effects of date, gall level, fertility level, and all possible interactions on chlorophyll content. No significant differences were detected in June 2020 at either site (Table 2.3). By July in Jonesboro and August in Washington, treatments without DAP applied had significantly lower chlorophyll content than treatments with DAP applied, regardless of gall presence or absence.

In 2021, there were significant effects of date, gall level, and their interaction on chlorophyll content at the Jonesboro site (Table 2.2). Again, significant differences were not detected until July. Leaf chlorophyll content was significantly greater in stems without galls than stems with galls at this site. At the Washington site, there were significant effects of date, gall level, fertility level, and the interaction between gall level and fertility level. In June, treatments without DAP applied had significantly lower chlorophyll content than treatments with DAP applied. By July, stems without galls or DAP applied had significantly lower chlorophyll content than other treatment combinations in Washington during July 2021 (Table 2.3).
Table 2.2. Chlorophyll content mixed model results as affected by gall level and fertilizer application during the wild blueberry growing season in Jonesboro, crop year in 2020 and prune year in 2021 as well as in Washington, prune year in 2020 and crop year in 2021. Letters indicate pairwise differences.

<table>
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<th>Chlorophyll Content Mixed Model Effect p-values</th>
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<td></td>
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<td>0.0323*</td>
<td>&lt;0.0001*</td>
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<td>0.0001*</td>
<td>0.0015*</td>
</tr>
<tr>
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<td>&lt;0.0001*</td>
</tr>
<tr>
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<td>0.3497</td>
<td>&lt;0.0001*</td>
</tr>
<tr>
<td>G x F x D</td>
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<td>0.2333</td>
<td>0.7292</td>
<td>0.0206*</td>
</tr>
</tbody>
</table>
Table 2.3. Chlorophyll content monthly means by gall level and fertility level combinations at the Jonesboro and Washington field sites during 2020 and 2021 field seasons. Measures were ceased in 2021 after the July harvest at the Washington site. Letters indicate pairwise differences.

<table>
<thead>
<tr>
<th>Year and Treatment</th>
<th>Chlorophyll Content (SPAD units)</th>
<th>Jonesboro</th>
<th>Washington</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>June</td>
<td>July</td>
</tr>
<tr>
<td><strong>2020</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Not Galled – No DAP</td>
<td>25.05</td>
<td>29.68b</td>
<td>27.02c</td>
</tr>
<tr>
<td>Not Galled + DAP</td>
<td>25.09</td>
<td>36.06a</td>
<td>33.75a</td>
</tr>
<tr>
<td>Galled – No DAP</td>
<td>24.59</td>
<td>28.54b</td>
<td>25.47c</td>
</tr>
<tr>
<td>Galled + DAP</td>
<td>26.31</td>
<td>35.12a</td>
<td>31.27b</td>
</tr>
<tr>
<td><strong>2021</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Not Galled – No DAP</td>
<td>33.52</td>
<td>38.06a</td>
<td>-</td>
</tr>
<tr>
<td>Not Galled + DAP</td>
<td>33.28</td>
<td>36.69ab</td>
<td>-</td>
</tr>
<tr>
<td>Galled – No DAP</td>
<td>33.72</td>
<td>35.17bc</td>
<td>-</td>
</tr>
<tr>
<td>Galled + DAP</td>
<td>34.60</td>
<td>34.31c</td>
<td>-</td>
</tr>
</tbody>
</table>
2.5 Discussion

Gall midge density was significantly greater where 493.18 kg/ha, a high rate, of DAP fertilizer was applied during both years at the Washington site and during the prune year (2021) at the Jonesboro site. There were also greater percent differences in gall midge density during prune years. This is consistent with previous findings where blueberry gall midge was detected in greater densities in prune than in crop fields, most likely due to the utilization of young leaf tissue by many gall-inducing insects for gall formation (Collins and Drummond, 2019; Rohfritsch, 1992). In 2020, the Jonesboro site was severely affected by leaf spot disease, which caused high rates of defoliation and may have impacted the non-significance of some results at that location. Further research is required to confirm gall midge preference toward plant tissue age and/or specific nutrients using choice and no-choice assays.

Galling reduced stem height and bud numbers at both sites. It also reduced the number of flowers, green fruit, and blue fruit per stem at the Washington site. These results agree with previous research by Collins and Drummond (2019). Future research should evaluate the effect of blueberry gall midge on the development rate of wild blueberry over more than one production cycle. Other examples of insect pest impacts on fruit development include delayed fruit ripening and uneven crop maturation caused by root-knot nematodes (*Meloidogyne hapla* and *M. incognita* Chitwood) in tomato (*Lycopersicon esculentum* Mill. ‘Veetbrite’) (Olthof and Potter, 1977; Reddy, 1985) and swede midges (*Contarinia nasturtii* Kieffer) in canola (*Brassica napus* L.) (Muzzatti et al. 2021).

DAP is a granular phosphorus and nitrogen heavy fertilizer used in acidic soil conditions and is the standard product used in wild blueberry production. DAP applications improved blueberry plant growth as expected (Percival et al. 2004; Yarborough and Smagula, 2013),
resulting in more buds per stem at both sites, and taller stems and greater number and weight of ripe berries per stem at the Washington site, despite increases in habitat availability (blueberry cover) and gall density. At the Washington site, yields in treatments where DAP was applied were significantly greater than in treatments without DAP applied even when galling was present. Ideally, wild blueberry productivity is best with sufficient nutrient availability and without galls. Applying DAP may therefore help compensate for the impact galls have on plant growth despite also providing more habitat and, in turn, more opportunity for gall midges residing in the blueberry canopy to feed and reproduce.

Plots where DAP was applied resulted in significantly greater soil nitrogen and phosphorus levels at the Washington site as well as significantly greater blueberry plant cover in the prune year at the Jonesboro site and both years at the Washington site. DAP application did not show a significant effect on foliar nutrient levels, but regression analysis did indicate a clear trend that gall density increased with increasing foliar nitrogen and phosphorus. Earlier foliar sampling of young tissue during the prune year may have been more valuable, since studies in other crops have shown leaf tissue nutrients to decrease with plant age (Bhangoo and Albritton, 1972; Pinkerton, 1991). Variance in the regression analysis may also be due to limiting factors such as sufficient moisture or protection from winter injury, which must be provided to achieve the full benefits of fertilizing wild blueberry (Yarborough and Smagula, 2013). Leaf chlorophyll content was significantly greater in stems located in fertilized plots during both years at the Washington site and during the crop year (2020) at the Jonesboro site. Significantly greater chlorophyll content indicated greater nitrogen levels in foliar tissues, a major nutrient required to support wild blueberry plant growth (Calderwood et al. 2020; Percival and Sanderson, 2004).

Physical differences observed in plants where DAP was applied included lighter and brighter
green coloration from new growth of leaves at shoot tips and therefore taller stems than those of plants where it was not applied during the first year at both sites (personal observation). These observations were demonstrated in the significant difference in stem height in Washington. Further research is needed regarding the uptake of fertilizer nutrients over time in wild blueberry with and without gall midge infestations. A study using multiple rates of DAP would be valuable to identify the level of DAP that would not significantly increase gall density, or a maximum level at which gall density stops increasing.

Our results agree with gall midge activity in cranberry, where greater abundances of this pest are more likely to be found in young or heavily fertilized cranberry beds with younger, nitrogen-rich leaves (Leduc and Turcotte, 2004). Similar trends are seen in other species of insects. Diamondback moths (Plutella xylostella (Lepidoptera: Plutellidae)) reared on Brassica oleracea showed a positive correlation in population growth with nitrogen fertilizer used in growing host plants (Fox et al. 1990). Increasing the amount of nitrogen supplied to wheat (Triticum aestivum) and barley (Hordeum vulgare) plants was positively correlated with increased performance in the rose-grain aphid (Metopolophium dirhodum (Hemiptera: Aphididae)) (Honek, 1991). Further research should follow gall midge infestation after fertilizer application in the prune year for more than one production cycle to record fertilizer effects on soil and plant nutrients. Research documenting larval fitness with and without fertilizer application would also be valuable in understanding generational success with this pest.

Collins and Drummond (2019) developed economic thresholds, or levels at which the costs of management and crop loss are equal, for this pest. For growers who receive average yields close to 4,453 kg/ha and who receive between $1.10 and $4.40/kg of blueberries, the economic threshold is between 1.5% and 5% infestation, with a lower and narrower threshold for
high-yielding growers receiving a higher price. This is based on an average crop loss of 50% due to galling. Crop loss in this study ranged between 31.41% and 85.25% based on the yield for fertilized stems without galls compared to other treatment combinations (Table 2.1). The average yield in 2020 was 2,567 kg/ha and average annual price was 1.33/kg (USDA NASS, 2021a), which was low compared to the previous five-year average. If we use the crop loss percentages measured in this study, the 2020 average yield and price received, and a proportionate injury reduction due to the control between 25% and 100% (to represent both low and high efficacy of control measures), the economic threshold ranges between 4.29% and 46.63% infestation. This was assuming the cost of management was $125/ha, the same cost used by Collins and Drummond (2019), not unusual for pest control costs in a conventional grower budget (Yarborough and D’Appollonio, 2017). No control measures were used in this study. However, growers who have fertilized and are considering control measures such as insecticides or early burning should wait until a greater percent infestation occurs, as the value of crop loss will be greater when low efficacy control tactics are used, low prices are received, or low yields are produced.

This research shows that it is important for farmers to consider whether DAP is necessary for crop production based on the level of gall midge infestation in their fields and their production goals. Reducing the amount of DAP applied may prevent increasing pest presence and environmental damage, such as soil nutrient leaching from fertilizer overuse. Applying fertilizer is only beneficial to increase yield in wild blueberry when other limiting factors such as pollination and soil moisture are sufficient (Yarborough and Smagula, 2013). Growers might also consider maintaining budgets that include fertilizer costs if blueberry gall midge is present in their fields rather than forgoing nutrient inputs when market prices are low. This is based on
our results that show yields were significantly greater where DAP was applied despite presence of galls. Growers should monitor fields for gall midge percent infestation and only apply nutrient inputs based on leaf nutrient levels. Soil nutrient levels are not always indicative of leaf nutrient levels and may be low when leaf nutrients are not (Yarborough and Smagula, 2013). Appropriate nutrient testing is required to reduce the unnecessary attraction of blueberry gall midge to wild blueberry while choosing the required nutrient inputs carefully. Our study suggests that blueberry gall midge is more attracted to areas where a high rate of DAP fertilizer has been applied, however, yields were still greater where this rate of fertilizer was used.
REFERENCES


APPENDICES

Appendix A: Chapter 1 Supplementary Information

Figure A.1. Weekly drought conditions in Maine per year by percent area of state affected. Adapted from United States Drought Monitor data for the National Integrated Drought Information System.

![Historical Drought Conditions in Maine](chart.png)
Appendix B: Chapter 2 Supplementary Information

Figure B.1. Example of one replicate of randomized complete block split-plot design
BIOGRAPHY OF THE AUTHOR

Becky Gumbrewicz was born in New Haven, Connecticut on August 6, 1997. She was raised in Oxford, Connecticut and graduated from Oxford High School in 2015. She attended the University of Rhode Island and graduated in 2019 with a Bachelor of Science degree in Environmental Science and Management. She moved to Maine and entered the Plant, Soil, and Environmental Sciences graduate program at the University of Maine in the spring of 2019. Becky is a candidate for the Master of Science degree in Plant, Soil, and Environmental Sciences from the University of Maine in December 2021.